Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
www.phytojournal.com
JPP 2020; 9(6): 541-545
Received: 15-08-2020
Accepted: 16-10-2020

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Effect of different biostimulants on growth parameters of maize in red soils of Karnataka

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Abstract

A field experiment was conducted to evaluate the performance of maize upon applicaton of different biostimulants namely, humic acid extracted from FYM, Spirulina algal extract and microbial consortia in red soils of zone 6 of Karnataka during *Kharif* 2018 on sandy loam soil at college of Agriculture, V.C. Farm, Mandya. The experiment was laid out in RCBD with thirteen treatments including control, 100% RDF, 100% RDF + microbial consortia (MC), 100% RDF + MC + humic acid @ 0.25 and 0.50%, 100% RPP + MC + algal extract @ 10 and 20% and the above treatments were repeated with 75% RPP. Results revealed that application of biostimulants had a significant effect on growth of maize. Foliar application of algal extract at 30 and 45 DAS was performing better than other treatments. Higher plant height, number of leaves, leaf area, SPAD reading and dry matter accumulation was recorded in the treatment receiving 100% RPP with microbial consortia and algal extract (T_6 and T_7) which might due to the effect of plant growth regulators present in the biostimulants.

Keywords: Biostimulant, Humic acid, Algal extract, Microbial consortia.

Introduction

Cultivation of high yielding varieties or hybrids to augment the food production has demanded the use of synthetic chemical fertilizers. The continuous use of fertilizers in the crop cultivation showed some deleterious effect on soil quality and environment. Thus maintenance of soil health is very important for the biochemical processes including the transformation of organic matter (Miltner *et al.*, 2011) [11], nutrient release (Wichern *et al.*, 2007) [25] and degradation of xenobiotics. Hence, agronomic technologies that takes care of soil health and at the same time protect the plant from biotic and abiotic stresses besides ensuring plant vigour, higher yield, robust root growth, improved nutrient uptake and enhanced microbial diversity is the need of the hour. Biostimulants in crop production is recently gaining lot of importance as the most sustainable and viable technology for the producers who are looking for the production of quality produce, without compromising the yield and at the same time without causing undue stress on soil resources and environment.

The European Biostimulants Industry Council (EBIC, 2016) [6] defined biostimulants as "Substances or microorganisms whose function when applied to plants or the rhizosphere is to stimulate natural processes to benefit nutrient uptake, nutrient use efficiency, tolerance to abiotic stress, and/or crop quality, independent of its nutrient content".

Biostimulants are either natural or synthetic organic substances containing hormones or precursors of plant hormones, when applied in lower concentration to soil or seed (seed coating) or plant (foliar spray) favours the growth of the plant by improving the vital physiological processes of the crop allowing higher yields and quality produce. The role of biostimulants in improving the yield and quality of the crop produce and soil properties are not clearly demonstrated as these are composed of number of components as ingredients.

The biostimulants used in this field experiment were humic acid extracted from FYM, Spirulina algal extract and microbial consortia.

Material and methods

Field experiment was conducted during *Kharif* 2018 on at College of Agriculture, Vishweshwaraiah Canal (V. C.) Farm, Mandya, Karnataka. The experiment consists of 13 treatment combinations as mentioned below

Treatment details

Treatment	Details	Treatment	Details
T_1	Control	T_8	75 % RPP
T_2	100 % RPP	T_9	75 % RPP + MC
T_3	100 % RPP + MC	T_{10}	75 % RPP + MC + HA 0.25 %
T_4	100 % RPP + MC + HA @ 0.25 %	T_{11}	75 % RPP + MC + HA 0.50 %
T_5	100 % RPP + MC + HA @ 0.50 %	T_{12}	75 % RPP + MC + AE @ 10 %
T_6	100 % RPP + MC + AE @ 10 %	T_{13}	75 % RPP + MC + AE @ 20 %
T_7	100 % RPP + MC + AE @ 20 %		

NOTE: MC- Microbial consortia HA- Humic acid AE- Algal extract RPP – Recommended package of practices as per the UAS B package of practices includes application of Recommended dose of NPK for Maize is 150:75:40 kg ha^{-1} + 10 kg ha^{-1} $ZnSO_4$, with farm yard manure (FYM) at the rate of 10 t ha^{-1}.

The microbial consortia was applied to soil along with FYM i.e. at 15 days before sowing of maize. While, humic acid and algal extract was foliar sprayed at 30 and 45 days after sowing.

Extraction of humic substances
Humic acid was extracted from well decomposed FYM by alkaline extraction method and further acidification as described by Stevenson (1981) [20]. Five kg of air dried FYM was weighed and transferred to plastic container to which 25 liters of 0.5 N NaOH was added and the contents were shaken for 24 hours (Schnitzer and Skinner, 1968) [18]. The dark coloured supernatant solution was separated by filtration and collected. Then the supernatant was acidified and centrifuged to obtain humic acid. Precipitation and centrifugation was repeated to attain partial purification of humic acid fraction. Then it was placed in oven and dried at 60 ˚C to a constant weight. The humic acid obtained was ground and diluted to get the required concentration.

Microbial consortia
Microbial consortia consisting of N- fixer + P- solubilizer + K- solubilizer + *Pseudomonas fluorescens* + *Trichoderma viridae* was obtained from Biofertilizer Unit, University of Agricultural Sciences, Bangalore and applied to soil along with FYM (15 days before sowing) at the rate of 2 kg per acre.

Production of the algal extract
The mother culture of *Spirulina platensis* was obtained from Center for Conservation and Utilization of Blue green Algae, IARI, New Delhi. Two ml of mother culture was inoculated into media broth to get sub-cultures for future use. Fifty ml culture was mixed initially with 500 ml zorrouck’s medium (pH 10). The culture was kept in an orbital shaker with natural illumination (3000 lux) and temperature of 30 °C for 7 days. Using the subcultures, the mass production has been carried out to obtain spirulina algal mass. The extract obtained was smashed in pestle and mortar and the solution was considered as 100 per cent. The solution was further diluted to get required concentration.

Table 1: Initial Physico-chemical properties of soil at the experimental site

Sl. No	Parameter	Method	Value
Physical Properties			
1	Sand (%)	International pipette method	80.51
2	Silt (%)		9.14
3	Clay (%)		9.23
4	Textural class		Sandy loam
5	Bulk density (Mg m^{-3})		1.51
Chemical properties			
1	pH(1:2.5)	Potentiometry	7.21
2	$EC_{2.5}$ (dS m^{-1})	Conductometry	0.17
3	Organic carbon (g kg^{-1})	Wet digestion	5.70
4	Available N (kg ha^{-1})	Alkaline potassium permanganate distillation method	276.87
5	Available P_2O_5 (kg ha^{-1})	Olsens extractant method, Colorimetry	35.33
6	Available K_2O (kg ha^{-1})	Ammonium acetate extractant method, Flame photometry	260.80
7	Available Ca (cmol (p$^+$) kg^{-1})	Ammonium acetate extractant method, Versenate titration method	4.71
8	Available Mg (cmol (p$^+$) kg^{-1})		3.13
9	Available S (mg kg^{-1})	$CaCl_2$ extraction, Turbidimetry	14.72
10	DTPA Fe (mg kg^{-1})	Atomic absorption spectrophotometry	12.76
11	DTPA Zn (mg kg^{-1})		0.79
12	DTPA Mn (mg kg^{-1})		7.94
13	DTPA Cu (mg kg^{-1})		0.61
14	Hot water soluble Boron (mg kg^{-1})	Hot water extraction method and colorimetry using Azomethine-H	0.51

The soil at the experimental site was sandy loam in texture with 80.51, 9.14, and 9.23 per cent sand, silt and clay, respectively and bulk density of soil was 1.51 Mg m^{-3}. The soil was neutral in reaction (pH 7.21) and low in soluble salts (0.17 dS m^{-1}). The soil was medium in organic carbon (5.70 g kg^{-1}), low in available nitrogen (276.87 kg ha^{-1}), medium in available P_2O_5 (35.33 kg ha^{-1}), medium in available K_2O (260.80 kg ha^{-1}) and sufficient in sulphur (14.72 mg kg^{-1}). The exchangeable calcium and magnesium content of soil was 4.71 and 3.13 c mol kg^{-1}, respectively. The content of DTPA extractable iron, zinc, manganese, copper and hot water soluble boron was 12.76, 0.79, 7.94, 0.61 and 0.51 mg kg^{-1}, respectively.

Results and discussion

Table 2: Plant height (cm) at different growth stages as affected by application of different biostimulants

Treatments	30 DAS	60 DAS	90 DAS	Harvest
T_1	22.10	130.10	136.10	136.90
T_2	31.10	181.60	190.60	191.60
T_3	30.80	184.30	194.30	194.90
T_4	31.40	190.10	195.10	196.10
T_5	32.60	192.80	197.80	198.80
T_6	31.50	196.00	205.77	206.37
T_7	30.90	202.43	213.13	214.63
T_8	28.90	173.10	179.20	182.00
T_9	28.50	175.40	181.90	182.40
T_{10}	27.80	179.10	182.20	183.10
T_{11}	27.10	178.40	183.90	185.30
T_{12}	29.50	184.03	190.53	192.73
T_{13}	28.80	185.23	191.43	193.57
S.Em±	1.25	7.40	7.12	7.13
CD @ 5%	3.64	21.60	20.77	20.81

Table 3: Number of leaves at different growth stages as affected by application of different biostimulants

Treatments	30 DAS	60 DAS	90 DAS	Harvest
T_1	5.17	8.71	8.97	8.11
T_2	7.53	13.17	13.57	10.51
T_3	7.60	13.23	13.63	10.30
T_4	7.81	13.57	13.77	10.90
T_5	7.75	13.91	14.52	11.57
T_6	7.71	14.31	14.73	11.85
T_7	7.85	14.77	15.31	12.11
T_8	6.33	11.10	12.03	10.11
T_9	6.57	11.67	12.93	10.40
T_{10}	6.20	12.15	12.95	10.33
T_{11}	6.50	12.87	13.01	10.81
T_{12}	6.31	13.01	13.53	10.95
T_{13}	6.47	13.37	13.97	11.00
S.Em±	0.31	0.56	0.59	0.49
CD @ 5%	0.90	1.65	1.72	1.42

Plant height

The plant height at different growth stages of crop as influenced by biostimulants application is indicated in Table 2.

At 30 DAS, plant height before foliar application of HA and AE is presented under Table 10 and the pooled analysis revealed that significantly higher plant height of 32.60 cm was recorded in T_5 (100% RPP + MC + HA 0.5%) which was on par with all other 100 per cent RPP treatments and significant with 75 percent RPP treatments with or without biostimulants. Significantly lower plant height of 22.10 cm was recorded in control (T_1).

The plant height at 60 DAS indicated that, higher plant height of 202.43 cm was recorded in T_7 which was on par with treatment having 100 per cent RPP with either of the biostimulant application (T_3, T_4, T_5 and T_6) and treatments receiving 75% RPP with algal extract (T_{12} (185.63 cm) and T_{13} (186.53 cm)) but was significant with the remaining treatments. Lower plant height was recorded in control (130.10 cm).

Plant height at 90 DAS and at harvest indicated that, significantly higher plant height of 213.13 and 214.63 cm, respectively was recorded in T_7 which was on par with treatments receiving 100% RPP with either of the biostimulant (T_3 to T_6) but significant with rest of the treatments. Lower plant height of 136.10 and 136.90 cm at 90 DAS and at harvest, respectively was registered in control.

Number of leaves

Number of leaves in the pooled analysis varied significantly due to application of biostimulants and indicated in Table 3.

At 30 DAS, higher number of leaves per plant was recorded in treatment having 100 per cent RPP + MC + 20 per cent AE (7.85) which was on par with all the 100 per cent RPP treatments (T_2 to T_6). Lower number of leaves were recorded in control (5.17).

At 60 DAS, 90 DAS and at harvest, higher number of leaves were recorded in T_7 (14.77, 15.31 and 12.11, respectively) which were on par with treatments receiving 100% RPP with biostimulants and 75% RPP treatments with algal extract (T_{12} and T_{13}). Lower number of leaves per plant was observed in T_1 (8.71, 8.97 and 8.11, respectively).

Table 4: Leaf area (cm^2) and dry matter acculation (g per plant) at different growth stages as affected by application of different biostimulants in maize

	Leaf area (cm^2)				Dry matter accumulation (g plant^{-1})			
Treatments	30 DAS	60 DAS	90 DAS	Harvest	30 DAS	60 DAS	90 DAS	Harvest
T_1	1873	3682	3723	3218	9.50	54.12	77.20	79.90
T_2	2311	5715	5764	5698	11.15	90.48	130.61	141.57
T_3	2538	5901	5923	5842	11.50	92.25	132.96	143.54
T_4	2390	5955	6011	5897	11.05	93.91	134.60	149.04
T_5	2431	6034	6065	5985	10.95	94.70	135.82	154.26
T_6	2497	6212	6274	6107	11.01	95.22	137.11	156.34
T_7	2364	6310	6374	6198	11.19	96.81	138.70	157.15
T_8	2116	5284	5314	5105	10.18	80.26	115.75	127.73
T_9	2198	5471	5512	5311	10.24	82.15	117.28	128.16
T_{10}	2219	5514	5568	5397	10.46	83.04	118.45	132.48
T_{11}	2265	5654	5691	5438	10.55	83.99	119.40	135.78
T_{12}	2286	5702	5769	5710	10.28	84.19	120.81	139.40
T_{13}	2302	5759	5842	5637	10.31	85.96	122.90	143.23
S.Em±	103.27	250.25	252.33	238.85	0.48	3.80	5.44	6.06
CD @ 5%	301.42	730.44	736.49	697.15	NS	11.08	15.87	17.68

Table 5: SPAD reading of maize as affected by application of different biostimulants

Treatments	First spray (30 DAS)		Second spray (45 DAS)	
	SPAD one day before spraying	SPAD five days after spraying	SPAD one day before spraying	SPAD five days after spraying
T_1	25.31	3682	3723	3218
T_2	38.93	5715	5764	5698
T_3	38.61	5901	5923	5842
T_4	39.90	5955	6011	5897
T_5	41.80	6034	6065	5985
T_6	42.60	6212	6274	6107
T_7	40.23	6310	6374	6198
T_8	34.30	5284	5314	5105
T_9	35.47	5471	5512	5311
T_{10}	35.73	5514	5568	5397
T_{11}	36.11	5654	5691	5438
T_{12}	37.25	5702	5769	5710
T_{13}	37.54	5759	5842	5637
S.Em±	1.64	250.25	252.33	238.85
CD @ 5%	4.79	730.44	736.49	697.15

Leaf area (cm^2)

Data at 30 DAS indicated that higher leaf area of 2538 cm^2 was recorded in T_3 (100% RPP + MC), which was on par with all other treatments except T_1 (1873 cm^2), T_8 (2116 cm^2), T_9 (2198 cm^2) and T_{10} (2219 cm^2) (Table 4).

At 60 and 90 DAS, higher leaf area of 6310 and 6374 cm^2, respectively was recorded in T_7 treatment (100% RPP + MC + 20% AE) which was significantly higher than T_1 (3682 and 3723 cm^2), T_8 (5284 and 5314 cm^2), T_9 (5471 and 5512 cm^2) and T_{10} (5514 and 5568 cm^2). Lower leaf area of 3682 and 3723 cm^2, respectively was recorded in control.

At harvest, higher leaf area of 6198 cm^2 was recorded in T_7 which was significantly higher than control (3218 cm^2), T_8 (5105 cm^2), T_9 (5311 cm^2), T_{10} (5397 cm^2) and T_{11} (5438 cm^2).

Dry matter production (g $plant^{-1}$)

Dry matter production in maize plants at different intervals of time is presented in Table 4.

At 30 DAS, dry matter accumulation did not varied significantly. However, higher value of 11.50 g per plant was recorded in T_3 (100% RPP + MC) and lower value of 9.50 g per plant was recorded in control.

At 60 DAS, 90 DAS and harvest, higher dry matter accumulation of 96.81, 138.70 and 157.15 g $plant^{-1}$, respectively was recorded in T_7 treatment which was on par with all other 100% RPP treatments and T_{13} treatment and significant with rest of the treatments.

SPAD readings

SPAD reading varied significantly at 30 and 45 DAS due to application of biostimulants in 2018, 2019 and in pooled analysis (Table 5).

At 30 DAS, higher SPAD reading of 42.60 was recorded in T_6 (100% RPP + MC + 10% AE) which was on par with all the 100% RPP treatments with soil application of microbial consortia (T_3 to T_7) and it was significant with control and all the treatments with 75% RPP. Lower SPAD reading was recorded in control (25.31). However, after foliar application of biostimulants, higher SPAD reading of 45.18 was recorded in T_7 which was on par with T4 (40.28), T_5 (42.58) and T_6 (44.41) and significant with rest of the treatments. Lower value of 25.41 was recorded in control.

At 45 DAS, before foliar application of biostimulants higher SPAD value of 53.21 was recorded in T7 which was on par with all the treatments with 100% RPP and T13 (47.48) and significant with rest of the treatments. Lower value of 33.38 was recorded in T_1 treatment. However, five days after application, SPAD reading increased to 55.70 in T_7 treatment (100% RPP + MC + 20% AE) which was significant with all other treatments except T5 (50.19) and T_6 (53.87). Control recorded lower value of 33.57.

Higher SPAD reading upon application of biostimulants (30 DAS onwards) might be due to increased chlorophyll content with graded levels of humic acid and algal extracts application along with NPK fertilizers + MC as a result of additional supply of N and Mg and efficient absorption of N and Mg from soil that are vital components of chlorophyll. The increase in chlorophyll content increases the photosynthetic efficiency of maize which ultimately results in better growth and yield of crop. Similar increase in chlorophyll content upon application of biostimulants were evidenced by Sure *et al.* (2012) [21] in cucumber; Arjumend *et al.* (2015) [2] in wheat; El-Ghamry *et al.* (2009) [7], Tejada *et al.* (2018), Mohamed *et al.* (2017) [12], Raphael *et al.* (2018) [13] and Elizabeth *et al.* (2019) [8] in maize.

Increased growth parameters with the application of biostimulants at different concentration might be due to improvement in the physiological functions, structural function and stimulation of plant vigour as these biostimulants were composed of number of amino acids and hormones. The cytokinin and auxin components of biostimulants are strong promoters of shoot and root growth by stimulating the cell division and differentiation. Besides, applied biostimulants have also provided some essential nutrient elements which are absorbed through leaves. Similar increase in the growth parameters upon foliar application of HA was reported in wheat, corn and barley by Chen and Aviad (1990) [3]; Delfinea *et al.* (2005) [5] in rice; Sharif *et al.* (2006) [19], Reza and Moghadam *et al.* (2014) [15] and Sagar *et al.*, (2020) [17] in corn and Kiran *et al.* (2020) in cowpea. The positive response of crops to application of algal extract have been reported by Colla (2017) [4] in tomato; Sylwia *et al.*, 2019 [22] and Rathore *et al.* (2009) [14] in soybean; Yaghoub *et al.* (2019) [26] in lettuce; Fatma *et al.* (2014) [9], Andrade *et al.* (2018) [1] and Safinaz and Ragaa (2013) [16] in maize; Szczepanek and Grzybowski (2016) [23] in wheat.

The data on growth parameters of maize *viz.* plant height, number of leaves per plant, leaf area and total dry matter production recorded in treatments receiving 75% RPP + soil application of microbial consortia + foliar application of 20% AE and 100% RPP alone were statistically on par with each other. These results clearly indicate that the extent of increase in growth parameters that could be obtained with 100 per cent RPP application is possible with application of 75 per cent RPP when biostimulants were applied. The statistically similar values recorded with 75 per cent RPP when applied with biostimulants to that of 100 per cent RPP might be improvement in shoot and root growth, plant vigour and nutrient absorption with the application of AE as biostimulant.

Conclusion

75% RPP treatments along with microbial consortia and algal extract is giving on par results with that of 100% RPP treatments which results in saving of 25% fertilizer input consequentially results in greater resource use efficiency. The above study clearly emphasised beneficial effects of biostimulants in improving the crop growth and also maintaining soil health by reducing the fertilizer usage.

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